UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
CHEMICAL SAFETY AND
POLLUTION PREVENTION

April 24, 2013

MEMORANDUM

Subject: Efficacy Review for EPA File Symbol 6836-GLI, Carbosan 50D
DP Barcode: 407822

From: Marcus Rindal, Microbiologist 4/24/13
Efficacy Evaluation Team
Product Science Branch
Antimicrobials Division (7510P)

Thru: Emily Mitchell, Chief
Product Science Branch
Antimicrobials Division (7510P)
4/24/13

To: Velma Noble PM31/Drusilla Copeland
Regulatory Management Branch I
Antimicrobials Division (7510P)

Applicant: Lonza, Inc.
Allendale
90 Boroline Road
Allendale, NJ 07401

Formulation from the Label:

Active Ingredient(s)	% by wt.
Didecyl dimethyl ammonium carbonate and Didecyl dimethyl ammonium bicarbonate	50.0%
Other Ingredients	50.0%
Total	100.0%

I. BACKGROUND

The product, Carbosan 50D (EPA File Symbol 6836-GLI), is a new product. The applicant requested to register the product as a broad spectrum disinfectant, food contact sanitizer, and deodorizer for use on hard, non-porous surfaces. The product is for use in residential, commercial, industrial, and institutional environments. The label claims that the one-step disinfectant product is also an effective food contact sanitizer in the presence of 500 ppm hard water. Studies were conducted at ATS Labs, located at 1285 Corporate Center Drive, Suite 110, in Eagan, MN 55121.

Carbosan 7.5D, Carbosan 20D and Carbosan 50D are 100% identical to Carbosan 7.5 (EPA Reg. No. 6836-332), Carbosan 20 (EPA Reg. No. 6836-331) and Carbosan 50 (EPA Reg. No. 6836-327) with the exception of disinfectant claims. Group A and B product chemistry, acute toxicology and sanitizing efficacy data are cited using the "selective-cite" method to support these registrations. Efficacy studies conducted on Carbosan 7.5 are included in this package to support all three products as broad-spectrum disinfectants.

This data package contained a letter from the applicant's representative (dated November 19, 2012), two studies (MRID 489944-01 and 489944-02), Statements of No Data Confidentiality Claims for both studies, and the proposed label.

II. USE DIRECTIONS

The dilutable spray product is designed as a one-step broad/general spectrum disinfectant and deodorizer, effective in the presence of 250 ppm hard water plus 5% organic serum. The product is designed as a sanitizer on previously cleaned food contact surfaces in the presence of 500 ppm hard water.

The product is designed for disinfecting and sanitizing hard, non-porous, food-contact and nonfood-contact inanimate surfaces in residential, commercial, institutional, and industrial environments such as those found in homes, kitchens, healthcare institutions, airports, boats, children's rooms, clinics, day care centers, dental offices, doctor's offices, health clubs, dorm rooms, hotels, motels, homes, laboratories, office buildings, physician's offices, rest stops, airplanes, bathrooms, beverage plants, cafeterias, cheese factories, convenience stores, dairy farms, drinking fountains, egg processing plants, fast food operations, fisheries, food processing plants, hotels, meat processing plants, milk processing plants, prisons, schools, USDA inspected food processing facilities, restaurants, and sick rooms. The label states the product may be used on surfaces such as: metal, chrome, enamel, stainless steel, glass, fiberglass, finished wood, fiberglass, glazed ceramic / enamel / porcelain, sealed granite / limestone / marble / slate / stone / terra cotta / terrazzo, linoleum, metal, vinyl, and plastic.

DISINFECTION: Dilute 1 part product to 853 parts water (3 oz. per 20 gallons of water). Apply diluted product to hard, nonporous surfaces, thoroughly wetting surfaces with a coarse trigger sprayer. Treated surfaces must remain wet for 5 minutes. Wipe dry with a cloth, sponge, or mop or allow to air dry. For heavily soiled areas pre-clean first. Spray 6-8 inches from the surface; rub with a brush, sponge, or cloth. Rinse all surfaces that come in contact with food with potable water before reuse. Do not use on utensils, glassware, and dishes as a disinfectant.

SANITIZATION: To sanitize food contact surfaces, dilute 1 part product to 20 parts water.

For sanitizing food processing equipment, dairy equipment, food utensils, dishes, silverware, glasses, sink tops, countertops, refrigerated storage and display equipment and other hard surfaces. Prior to application, remove gross food particles and soil by a pre-flush, or pre-scrape and, when necessary, pre-soak. Then thoroughly wash or flush objects with a good detergent or compatible cleaner, followed by a potable water rinse before application of the sanitizing solution. Apply diluted product to the pre-cleaned hard surface, with a cloth, mop, sponge, [coarse] sprayer or by immersion, thoroughly wetting surfaces. Surfaces must remain wet for at least [one minute] [60 seconds] followed by adequate draining [and air drying]. Do not rinse. Prepare fresh solution daily or more often if the use solution becomes diluted or soiled. For mechanical applications, use-solution may not be reused for sanitizing applications but may be reused for other purposes such as cleaning. Apply to sink tops, counter tops, refrigerated storage and display equipment and other stationary surfaces by cloth, sponge [or] brush [or coarse spray]. Immerse pre-cleaned glassware, dishes, silverware, cooking utensils and other similar size food processing equipment in diluted product for at least [one minute] [60 seconds]. Drain thoroughly and allow to air dry before reuse. Do not rinse.

III. AGENCY STANDARDS FOR PROPOSED CLAIMS

Broad/General Spectrum Disinfectants for Use on Hard Non-Porous Surfaces: The effectiveness of broad spectrum disinfectants (represented in labeling as having efficacy against both Gram-negative and Gram-positive bacteria) for use on hard surfaces must be substantiated by data derived using the AOAC Use-Dilution Method (for water soluble powders and liquid products) or the AOAC Germicidal Spray Products as Disinfectants Method (for spray products). Sixty carriers must be tested with each of 3 product samples, representing 3 different product lots, one of which is at least 60 days old, against both *Salmonella enterica* (formerly known as *Salmonella choleraesuis*) (ATCC 10708) and *Staphylococcus aureus* (ATCC 6538). To support products labeled as "disinfectants," killing on 59 out of 60 carriers is required to provide effectiveness at the 95% confidence level. If the product is intended to be represented as bactericidal in the presence of organic soil (one-step), an appropriate organic soil, such as 5 percent blood serum, should be included with the bacterial inoculum. For the AOAC International Use-Dilution Methods, the Germicidal Spray Products as Disinfectants test, and single-use towelettes, the product should kill the test microorganisms on 59 out of each set of 60 carriers/slides in ≤ten minutes. In addition, per the 2009 AOAC revisions for the Use-Dilution Method, the mean log density for *S. aureus* is to be at least 6.0 (corresponding to a geometric mean density of $1.0x10^6$); a mean log density <6.0 invalidates the test. The mean log density for *Salmonella enterica* is to be at least 4.0 (corresponding to a geometric mean density of $1.0x10^4$); a mean log density <4.0 invalidates the test.

Disinfectants for Use on Hard Surfaces (Additional Bacteria): Effectiveness of disinfectants against specific bacteria other than those named in the AOAC Use-Dilution Method, AOAC Germicidal Spray Products as Disinfectants Method, AOAC Fungicidal Test, and AOAC Tuberculocidal Activity Method, must be determined by either the AOAC Use-Dilution Method or the AOAC Germicidal Spray Products as Disinfectants Method. Ten carriers must be tested against each specific microorganism with each of 2 product samples, representing 2 different product lots. To support products labeled as "disinfectants" for specific bacteria (other than those bacteria named in the above test methods), killing of the specific microorganism on all carriers is required. In addition, plate count data must be submitted for each microorganism to demonstrate that a concentration of at least 10^4 microorganisms survived the carrier-drying step. These Agency standards are presented in DIS/TSS-1.

IV. SUMMARY OF SUBMITTED STUDIES

1. MRID 489944-01: "AOAC Germicidal Spray Method," Test Organisms: *Salmonella enterica* (ATCC 10708); *Staphylococcus aureus* (ATCC 6538), for Carbosan 7.5 (EPA Reg. No. 6836-332), by Nicole Albert. Study conducted at ATS Labs. Study completion date – June 23, 2011. Project Number A11517.

This study was conducted against *Staphylococcus aureus* (ATCC 6538) and *Salmonella enterica* (ATCC 10708). Three lots (Lot # 5893-028B, 5893-028D, 5893-028F) of the product (Lot # 5893-028B ≥60 days old), Carbosan 7.5, were tested using ATS Labs protocol # LZ01051911.GS.1 (copy provided). The test solution was prepared using 6.0 mL of the test substance and 762 mL of 250 ppm AOAC Synthetic Hard water (1:128 dilution). The prepared test substance was used within three hours of preparation. Fetal bovine serum was added to each culture to achieve a 5% organic soil load. Sixty (60) glass slide carriers per product lot per microorganism were inoculated with 10 µL of a 48-54 hour suspension of test organisms incubated at 35-37°C. For *Salmonella enterica*, the slides were allowed to dry at 36.1°C for 30 minutes with a 60% relative humidity. For *Staphylococcus aureus*, the slides were allowed to dry for 38 minutes at 36.0-36.1°C and at a 42-60% relative humidity. Each carrier in a horizontal position at staggered intervals was sprayed (3-4 sprays) with the test substance at a distance of 6-8 inches from the carrier surface. Each carrier remained in contact with the test substance for 5 minutes at 20-22°C and 37-47% relative humidity. Following exposure, the excess liquid was drained off the carrier. Individual carriers were transferred to 20 mL of Letheen Broth containing 0.07% Lecithin + 0.5% Tween 80 for neutralization and subculturing. All subcultures were incubated for 48±2 hours at 35-37°C. Following incubation, the subcultures were visually examined for the presence or absence of visible growth. For the dried colony counts, carrier/neutralizing broth mixtures were vortex mixed. Controls included those for purity, sterility, viability, neutralization confirmation, and carrier population.

2. MRID 489944-02: "AOAC Germicidal Spray Method," Test Organism: *Salmonella enterica* (ATCC 10708); *Escherichia coli* O157:H7 (ATCC 35150), for Carbosan 7.5 (EPA Reg. No. 6836-332), by Nicole Albert. Study conducted at ATS Labs. Study completion date – June 10, 2011. Project Number A11518.

This study was conducted against *Escherichia coli* O157:H7 (ATCC 35150). Two lots (Lot # 5893-028B and 5893-028D) of the product (Lot # 5893-028B ≥60 days old), Carbosan 7.5, were tested using ATS Labs protocol # LZ01051911.GS.2 (copy provided). The test solution was prepared using 2.00 mL of the test substance and 254.0 mL of 250 ppm AOAC Synthetic Hard water (1:128 dilution). The prepared test substance was used within three hours of preparation. Fetal bovine serum was added to each culture to achieve a 5% organic soil load. Ten (10) glass slide carriers per product lot were inoculated with 10 µL of a 48-54 hour suspension of test organisms incubated at 35-37°C. The slides were allowed to dry at 36.1°C for 30 minutes with a 60% relative humidity. Each carrier in a horizontal position at staggered intervals was sprayed (3-4 sprays) with the test substance at a distance of 6-8 inches from the carrier surface. Each carrier remained in contact with the test substance for 5 minutes at 21°C and 37% relative humidity. Following exposure, the excess liquid was drained off the carrier. Individual carriers were transferred to 20 mL of Letheen Broth containing 0.07% Lecithin + 0.5% Tween 80 for neutralization and subculturing. All subcultures were incubated for 48±2 hours at 35-37°C. Following incubation, the subcultures were visually examined for the presence or absence of visible growth. For the dried colony counts, carrier/neutralizing broth mixtures were vortex mixed. Controls included those for purity, sterility, viability, neutralization confirmation,

and carrier population.

V. RESULTS

Table 1. AOAC Germicidal Spray Products Test Method Results for Carbosan 7.5
Broad/General Spectrum Disinfectant Claim 5 min. Contact Time

MRID Number	Sample Batch Reference Number		Organism		Average Carrier Population (CFU/ carrier)
			S. aureus	*S. enterica*	
489944-01	Test Date 5/27/11	5893-028B	2/60	0/60	*S.a.* 2.8×10^6 *S.e.* 1.5×10^4
		5893-028D	0/60	0/60	
		5893-028F	0/60	0/60	
	Repeat Testing of Lot 5893-028B against *S. aureus**				
	Test Date 6/10/11	5893-028B	0/60	--	*S.a.* 2.8×10^6

* A study protocol Deviation (incorrectly identified as an Amendment) allowed for the repeat testing of Lot 5893-028B against *S. aureus* "for the evaluation of potential false positives [*sic*]."

Table 2. AOAC Germicidal Spray Products Test Method Results for Carbosan 7.5
Disinfectant Claim – Additional Organism 5 min. Contact Time

MRID Number	Sample Lot Number	Organism	Average Carrier Population (CFU/ carrier)
		Escherichia coli O157:H7	
489944-02	5893-028B	0/10	1.0×10^5
	5893-028D	0/10	

VI. CONCLUSIONS

1. The submitted efficacy data indicate that Lot 5893-028B failed to meet the necessary criteria for killing (≥59 out of 60 carrier sets) for *S. aureus*. All test control results indicate the testing was valid and support the failing results for Lot 5893-028B against *S. aureus* on the original test date, 5/27/11. In the absence of compelling justification for invalidating a portion of the original test, the repeat testing cannot be accepted. The submitted efficacy data DO NOT support the use of the product, Carbosan 50D, as a general or broad spectrum disinfectant with bactericidal activity (see Table 1.) against the following microorganisms on hard, non-porous surfaces with a contact time of 5 minutes in the presence of 5% organic soil:

   Staphylococcus aureus MRID 489944-01
   Salmonella enterica MRID 489944-01

   Purity controls were reported as pure. Viability controls were positive for growth. Sterility controls did not show growth. Neutralization confirmation results were acceptable.

2. The submitted efficacy data (MRID 489944-02) support the use of the product, Carbosan 50D, as a one-step broad or general spectrum disinfectant with bactericidal activity (see

Table 2.) against *Escherichia coli* O157:H7 on hard, non-porous surfaces with a contact time of 5 minutes, prepared with 250 ppm hard water, and in the presence of 5% organic soil. Purity controls were reported as pure. Viability controls were positive for growth. Sterility controls did not show growth. Neutralization confirmation results were acceptable.

VII RECOMMENDATIONS

1. The proposed label claims are unacceptable regarding the use of the product, Carbosan 50D, as a one-step general or broad spectrum disinfectant. Testing of Lot 5893-028B against *Staphylococcus aureus* (ATCC 6538) failed to achieve the necessary killing (≥59 out of 60 carrier sets) for a 5 minute contact time prepared with 250 ppm hard water, and in the presence of 5% organic soil.

2. The proposed label claims are acceptable regarding the use of the product, Carbosan 50D, as a one-step general or broad spectrum disinfectant against *Salmonella enterica* (ATCC 10708). The necessary killing (10 out of 10 carrier sets) was observed for a 5 minute contact time prepared with 250 ppm hard water, and in the presence of 5% organic soil.

3. Page 9 of proposed label: Verify last dilution in the box is accurate and formatted similarly to the other dilution directions.

4. Page 12 of proposed label: Remove all references to FOGGING from the label as no data were submitted to support this claim.